

Bacterial Stains

Fluorescent stains, conjugates & viability assays

Robust Dead Cell Staining Across the Spectrum

BactoView™ Dead Stains are unique DNA binding dyes designed for highly selective staining of dead bacteria for microscopy or flow cytometry analysis. Unlike traditional vital nucleic acid dyes like propidium iodide or ethidium homodimer that are excluded from live gram-negative bacteria but are able to penetrate live gram-positive bacteria, BactoView™ Dead Stains are excluded from both live gram-positive and gram-negative strains for highly selective live/dead discrimination. In addition to staining dead bacteria, BactoView™ Dead Stains also stain *Bacillus subtilis* endospores, but with dimmer fluorescence.

BactoView™ Dead staining is simple and does not require a wash step. The stains are available with a wide selection of emission wavelengths ranging from green to near-infrared, for microscopy or flow cytometry analysis (Fig. 1).

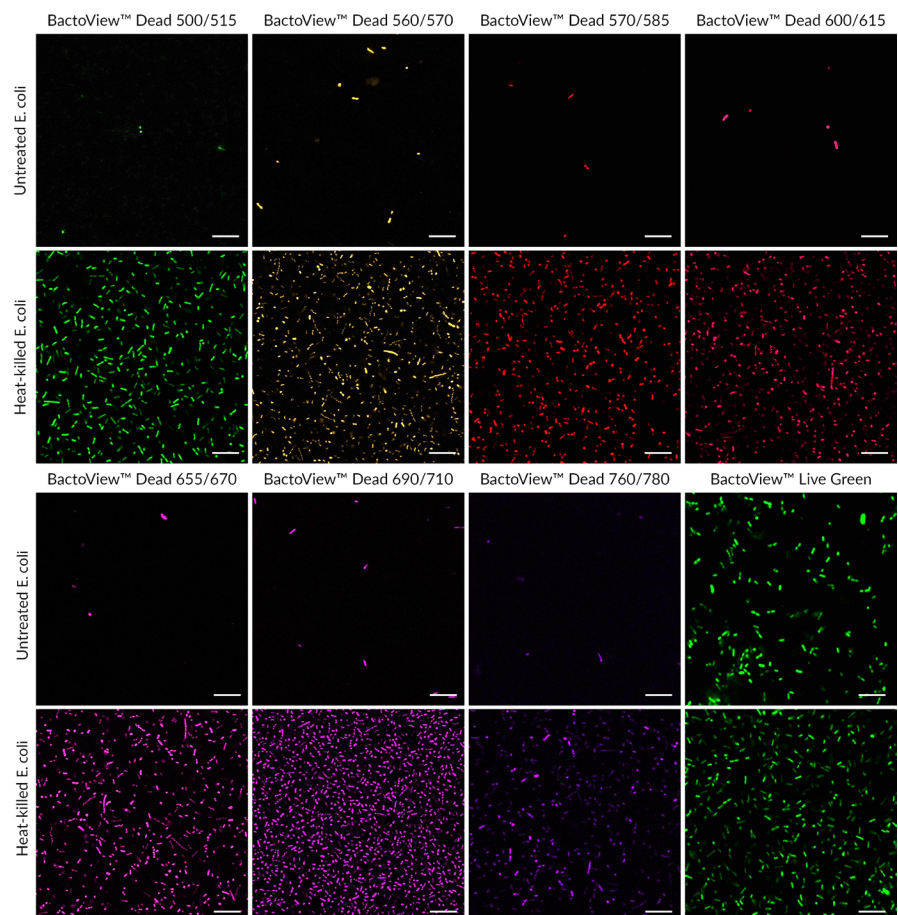
BactoView™ Dead Stain Features

- Vibrant staining of dead cells with minimal background
- Dead cell selective in gram-positive and gram-negative strains
- No-wash staining
- 7 colors from green to near-IR

Other tools for bacteria:

- 2- Color viability kits
- Live cell stains
- Gram stains
- Membrane/cell wall stains

Figure 1. Untreated or heat-killed *E. coli* were stained with BactoView™ Dead dye then imaged by confocal microscopy without a wash step. BactoView™ Live Green panels (far right) are representative of the typical cell densities of untreated and heat-killed *E. coli* samples. Scale bars: 20 um.



BactoView™ Live Fluorescent Bacterial Stains

BactoView™ Live Fluorescent Bacterial Stains were designed to stain both live and dead bacteria in a wide variety of gram-positive and gram-negative strains. The stains are cell-permeant, fluorogenic DNA stains and are suitable for microscopy or flow cytometry.

BactoView™ Live Stain Features

- Broad staining of live and dead bacteria
- Works with gram-positive and gram-negative bacteria
- Available with green or red fluorescence
- Suitable for microscopy or flow cytometry

Bright Staining of All Bacteria in Your Sample

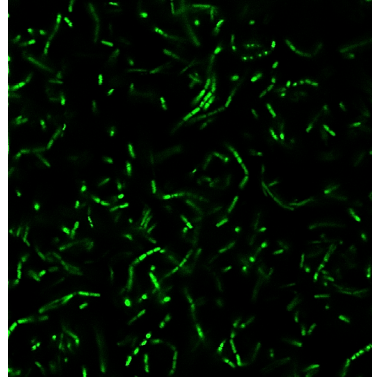


Figure 2. Live *B. subtilis* stained with BactoView™ Live Green, imaged by confocal microscopy.

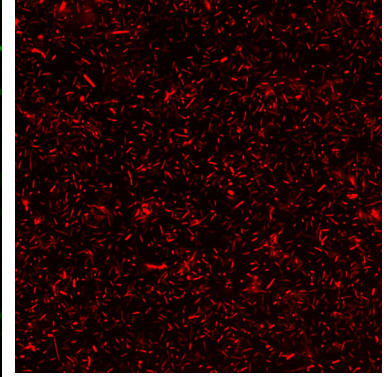


Figure 3. Live *E. coli* stained with BactoView™ Live Red, imaged by confocal microscopy.

BactoView™ Viability Kits

Robust Viability Assessment for Gram-Positive & Gram-Negative Strains

BactoView™ Viability Kits include Biotium's novel BactoView™ Stains for two-color staining of live and dead bacteria. BactoView™ Dead Stains are novel membrane-impermeant DNA binding dyes that selectively stain dead bacteria with compromised cell membranes. BactoView™ Viability Green Counterstain labels both live and dead bacteria with green fluorescence and is formulated for two-color staining with BactoView™ Dead (Fig. 4). Unlike many dead cell stains, BactoView™ Dead stains are efficiently excluded from gram-positive strains. The kits are available with red/green or far-red/green dye combinations.

Biotium also offers a Viability/Cytotoxicity Assay Kit for Bacteria Live & Dead Cells (Cat. No. 30027) which offers the best signal-to-noise for gram-negative strains. The kit provides two-color fluorescence staining of both live bacteria (green) and dead bacteria (red) using two DNA dyes, DMAO and EthD-III.

BactoView™ Viability Kit Features

- Suitable for gram-positive or gram-negative strains
- **BactoView™ Dead** is highly selective for dead cells (available with red or far-red fluorescence)
- **BactoView™ Viability Green Counterstain** labels both live and dead bacteria
- Rapid, no-wash staining

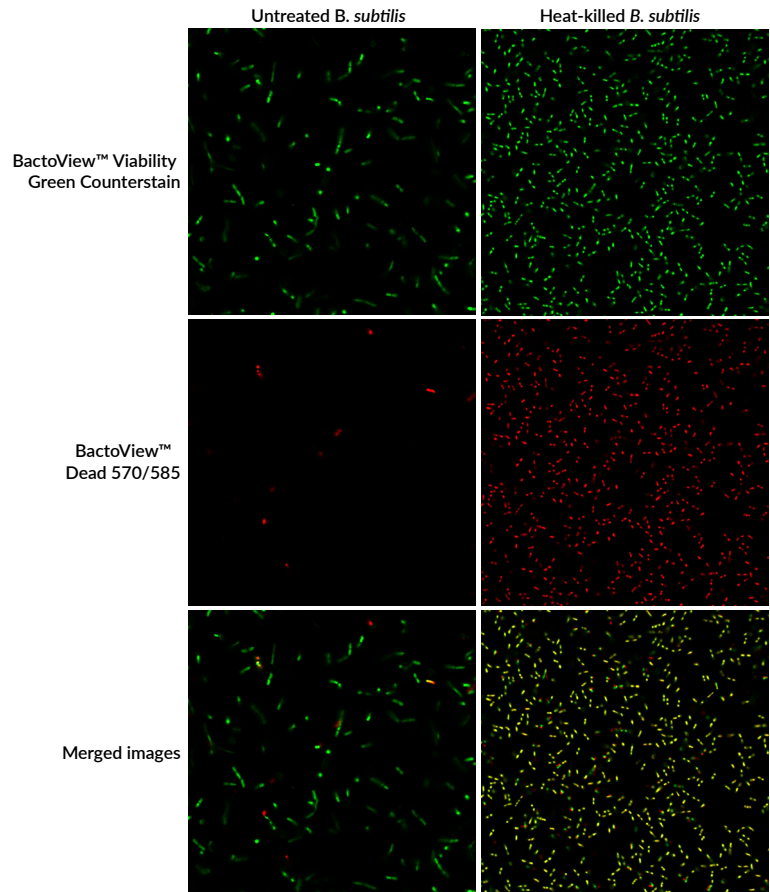


Figure 4. Live or heat-killed cultures of *B. subtilis* stained with the BactoView™ Viability Green/Red kit and imaged by confocal microscopy. Dead cells are stained red with BactoView™ Dead 570/585, all cells are stained green with BactoView™ Green Viability Counterstain. Co-stained cells appear yellow in the merged images.

BactoSpore™ Bacterial Stains

Bright and Sensitive Staining of Bacterial Endospores

Bacterial endospores are tough, dormant structures formed by certain strains of bacteria including *Bacillus* and *Clostridium* species in response to nutrient deprivation and other stressors. Endospores provide a reservoir of potentially infectious bacteria that are resistant to decontamination treatments. In addition, the spore coat is resistant to staining with bacterial detection reagents, making them difficult to study.

BactoSpore™ stains were developed to tackle the challenge of endospore detection by offering bright staining of endospores as well as live and dead bacteria. BactoSpore™ 488/540 Nucleic Acid Stain is a yellow fluorescent nucleic acid dye that is detectable in both the FITC channel and the PE channel for flow cytometry (Fig. 5). BactoSpore™ 485/500 Membrane Stain is a green

fluorescent lipophilic membrane dye for the FITC channel (Fig. 6).

BactoSpore™ Stain Features

- Bright staining of bacterial endospores
- Also stains live and dead bacteria
- For gram-positive and gram-negative strains
- Validated for microscopy and flow cytometry
- Options for staining membranes or DNA

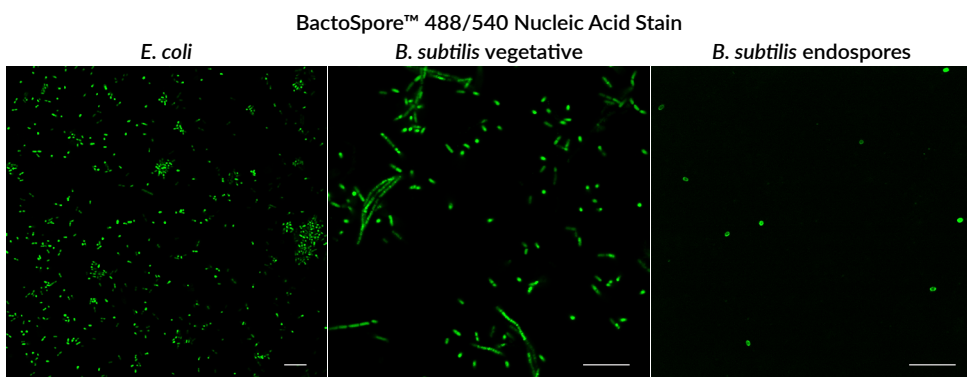


Figure 5. *E. coli*, vegetative *B. subtilis*, and *B. subtilis* endospores were stained with BactoSpore™ 488/540 Nucleic Acid Stain, then imaged by confocal microscopy in the FITC channel. Scale bars: 10 μm.

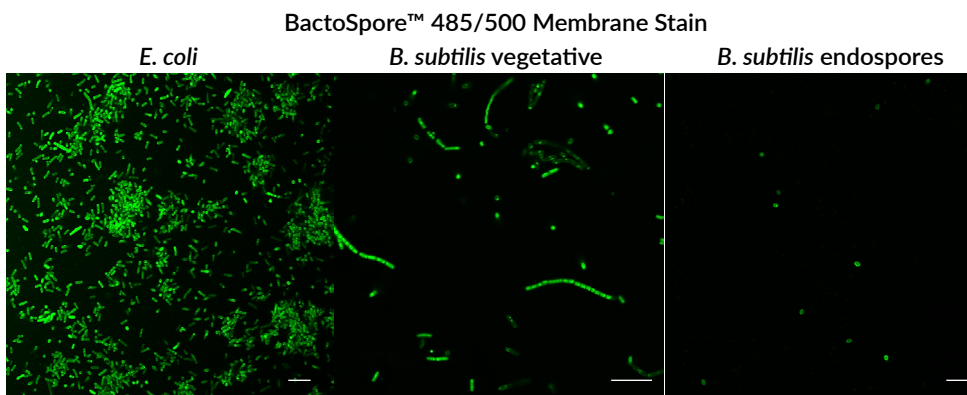


Figure 6. *E. coli*, vegetative *B. subtilis*, and *B. subtilis* endospores were stained with BactoSpore™ 485/500 Membrane Stain, then imaged by confocal microscopy in the FITC channel. Scale bars: 10 μm.

Bacterial Gram Stains & Kits

Biotium's wheat germ agglutinin (WGA) CF® Dye conjugates offer rapid and sensitive staining of gram-positive bacteria. The WGA conjugates are available with 13 color options as well as combination kits that include counterstains for viability analysis or all bacteria.

The Live Bacterial Gram Stain Kit (Cat. No. 32000) includes CF®594 WGA and DAPI for staining gram-positive cells red and all cells blue. The Bacterial Viability and Gram Stain Kit (Cat. No. 32001) includes CF®488A WGA, DAPI, and Ethidium Homodimer III (EthD-III) for labeling dead cells (Fig. 8).

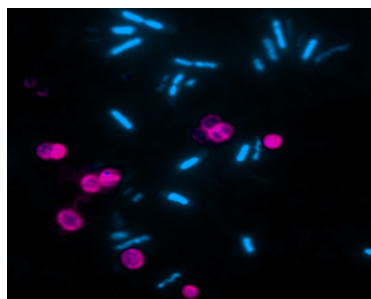


Figure 7. *E. coli* and *Staphylococcus* stained with CF®633 WGA (magenta) and DAPI (blue).

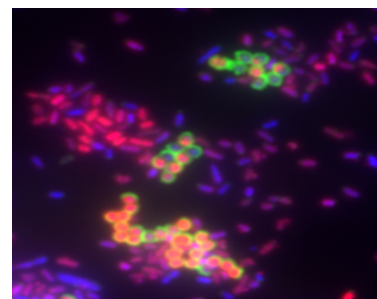


Figure 8. Heat-killed *Pseudomonas* and *Micrococcus* stained using the Bacterial Viability and Gram Stain Kit: CF®488A WGA (green), EthD-III (red), and DAPI (blue).

BactoView™ Live Fluorescent Bacterial Stains

Cat. No.	Product	Ex/Em (nm)	Detection Channel
40102	BactoView™ Live Green	500/520	FITC
40108	BactoView™ Live Red	572/675	Cy*3 (microscopy) PE-Cy*5 (flow cytometry)

BactoView™ Dead Stains

Cat. No.	Product	Ex/Em (nm)	Detection Channel
40107	BactoView™ Dead 500/515	497/515	FITC
40108	BactoView™ Dead 560/570	559/570	Rhodamine, PI, PE
40109	BactoView™ Dead 570/585	572/583	Rhodamine, PI, PE
40110	BactoView™ Dead 600/615	603/613	Texas Red® or PE-Texas® Red
40111	BactoView™ Dead 655/670	653/671	Cy*5, APC
40112	BactoView™ Dead 690/710	554/568	Cy*5.5
40113	BactoView™ Dead 760/780	683/707	Cy*7, APC-Cy*7

BactoSpore™ Bacterial Stains

Cat. No.	Product	Ex/Em (nm)	Detection Channel
40119	BactoSpore™ 485/500 Membrane Stain, 500X in EtOH	484/504	FITC
40120	BactoSpore™ 488/540 Nucleic Acid Stain, 500X in DMSO	488/536 (with DNA)	FITC or PE Channel

CF® Dye Wheat Germ Agglutinin (WGA) Conjugates

Cat. No.	Product	Ex/Em (nm)
29021	CF®350 Wheat Germ Agglutinin (WGA) Conjugate	355/450
29027	CF®405S Wheat Germ Agglutinin (WGA) Conjugate	411/431
29028	CF®405M Wheat Germ Agglutinin (WGA) Conjugate	416/452
29022	CF®488A Wheat Germ Agglutinin (WGA) Conjugate	490/516
29064	CF®532 Wheat Germ Agglutinin (WGA) Conjugate	531/552
29076	CF®555 Wheat Germ Agglutinin (WGA) Conjugate	554/568
29077	CF®568 Wheat Germ Agglutinin (WGA) Conjugate	562/584
29023	CF®594 Wheat Germ Agglutinin (WGA) Conjugate	593/615
29024	CF®633 Wheat Germ Agglutinin (WGA) Conjugate	629/650
29026	CF®640R Wheat Germ Agglutinin (WGA) Conjugate	642/663
29029	CF®680 Wheat Germ Agglutinin (WGA) Conjugate	681/698
29025	CF®680R Wheat Germ Agglutinin (WGA) Conjugate	680/701
29059	CF®770 Wheat Germ Agglutinin (WGA) Conjugate	770/797

Bacterial Viability Kits

Cat. No.	Product	Ex/Em (nm)	Detection Channel
32019	BactoView™ Viability Kit (Green/Red)	Green (498/522)/ Red (572/583)	FITC (Green)/ Rhodamine, PI, PE (Red)
32020	BactoView™ Viability Kit (Green/Far-Red)	Green (498/522)/ Far-Red (653/671)	FITC (Green)/ Cy*5, APC (Far-Red)
30027	Viability/Cytotoxicity Assay Kit for Bacteria Live & Dead Cells*	Green (496/528) Red (279, 532/625)	FITC (Green)/ Texas Red®, Cy*3 (Red)

*Not recommended for live/dead discrimination in gram-positive strains.

Other Stains for Bacterial Viability

Cat. No.	Product	Features
40017	Propidium Iodide (PI)*	
40014	Ethidium Homodimer I (EthD-I)*	<ul style="list-style-type: none"> Nuclear-specific dead cell stains For microscopy or flow cytometry
40051	Ethidium Homodimer I (EthD-III)*	
40061	RedDot™2 Far-Red Nuclear Stain	
32002... 32018	Live-or-Dye™ Fixable Viability Stains	<ul style="list-style-type: none"> Fixable dead cell stains Wide color selection For microscopy or flow cytometry
32010	Live-or-Dye NucFix™ Red Staining Kit	<ul style="list-style-type: none"> Fixable, DNA-binding dead cell stain For gram-negative bacteria only
10063	5-Cyano-2,3-ditolyl tetrazolium chloride (CTC)	<ul style="list-style-type: none"> Substrate for measuring bacteria respiratory activity Reduced to insoluble red formazon

*PI, EthD-I, and EthD-III are not recommended for live/dead discrimination in gram-positive strains.

Bacterial Gram Staining Kits

Cat. No.	Product	Ex/Em (nm)
32000	Live Bacterial Gram Stain Kit	CF®594 WGA: 593/614 DAPI: 358/461
32001	Bacterial Viability and Gram Stain Kit	CF®488A: 490/515 EthD-III: 532/625 DAPI: 358/461

Texas Red is a registered trademark of Thermo Fisher Scientific; Cy Dye is a registered trademark of Cytiva; Difco is a registered trademark of Becton, Dickinson and Company.